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Cannabinoid concentrations in blood and urine after smoking cannabidiol joints

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ABSTRACT

In Switzerland, the sale of cannabis with tetrahydrocannabinol (THC) content less than 1% has recently been legalized. As a consequence, cannabis with low THC and high cannabidiol (CBD) values up to approximately 25% is legally available on the market. In this study, we investigated cannabinoid blood and urine concentrations of a naive user and of a modeled chronic user after smoking a single CBD joint. Chronic use was modeled as smoking 2 joints per day for 10 days. Joints contained 200 mg of cannabis with THC concentrations of 0.94% and 0.8% and CBD concentrations of 23.5% and 17% in the naive-smoker and chronic-smoker experiment, respectively. After smoking, blood and urine samples were collected for 4 and 20 h after smoking start, respectively. THC blood concentrations reached 2.7 and 4.5 ng/mL in the naive and chronic user, respectively. In both cases, the blood THC concentration is significantly above the Swiss road traffic threshold of 1.5 ng/mL. Consequently, the user was legally unfit to drive directly after smoking. CBD blood concentrations of 45.7 and 82.6 ng/mL were reached for the naive and chronic user, respectively. During the 10-day smoking period, blood and urine samples were regularly collected. No accumulation of any cannabinoid was found in the blood during this time. Urinary 11-nor-9-carboxy-THC concentrations seemed to increase during the 10-day period, which is important in abstinence testing.

1. Introduction

Recently, there has been an increasing interest in cannabinoids for medical applications. Of the many phytocannabinoids, cannabidiol (CBD) has lately been the target of much research as it is non-intoxicating and has potential positive effects on various diseases such as cancer [1,2], epilepsy [3–5] and neurological diseases such as Parkinson's or Alzheimer's disease [6]. CBD has been reported to possess mild sedative effects while reducing negative delta-9-tetrahydrocannabinol (THC) effects such as anxiety [7]. The alteration of the THC-pharmacodynamics has been assigned to negative allosteric modulation of the CB_1-receptor [8]. For a review of potential therapeutic applications of CBD we refer to Pisanti et al. [9]

The consumption, sale and cultivation of cannabis sativa are forbidden or strictly regulated, in most countries [10]. According to the 2017 UN World Drug Report, cannabis is the most consumed illicit drug in the world, with an estimated 183 million users [10]. In the European Union (EU) and Switzerland, the classification of hemp as fiber or drug hemp is linked to its THC content. Swiss law considers hemp or hemp products to be drugs of abuse if the total concentration of tetrahydrocannabinol (THC) and tetrahydrocannabinolic acids (THCA) in the plant material or product is equal or higher than 1% (Appendix A, BetmVV-EDI) [11]. In the EU, the threshold is 0.2% [12].

In Switzerland, the sale of non-medical products containing cannabidiol has seen a massive increase in the past year. Hemp plants containing less than 1% THC and ca. 3–20% CBD are being sold in Switzerland for smoking as a tobacco replacement, as lifestyle products or with no declared use [13]. Although it is legal to produce, sell, possess and to consume these products, the Swiss Federal Office of Public Health strongly recommends that people consuming these products do not participate in traffic [13]. In principle, there is a zero-tolerance policy towards any drug of abuse while driving in Switzerland. In practice, the relevant ordinance of the Swiss Federal Roads Office [14] considers drivers with blood THC concentrations exceeding 1.5 ng/mL as momentarily unfit to drive. To this, a harmonized imprecision of ±30% is applied, giving a cut-off of 2.2 ng/mL. Although 1% THC is a low concentration in plant material, it could be enough to achieve blood concentrations above 2.2 ng/mL. Since the first appearance of CBD-rich products (hereinafter referred to as CBD) on the Swiss market, consumption of such products is often claimed as causative as to the THC concentrations detected in blood.

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This single case study, conducted as a self-experiment by one of the authors, aimed to investigate whether smoking a CBD joint is enough to achieve THC blood concentrations above the cut-off for driving in Switzerland in both single use (drug-naive) and in simulated chronic use. The results of this study are of particular importance in the interpretation of cases in which the driver claims to have only consumed CBD joints.

2. Materials and methods

2.1. Chemicals and preparation of stock solutions

THC, 11-nor-9-carboxy-delta-9-tetrahydrocannabinol (THC–COOH), 11-hydroxy-delta-9-tetrahydrocannabinol (THC–OH), CBD and cannabinol (CBN) at 1 mg/mL and THC-d3, THC–COOH-d9, THC–OH-d3, CBD-d3 and CBN-d3 at 0.1 mg/mL were purchased from Lipomed AG (Arlesheim, Switzerland). A reference mix containing 0.2 ng/μL CBN/CBD, 0.5 ng/μL THC/THC–OH and 5.0 ng/μL THC–COOH in methanol (MeOH) was prepared. An internal standard mix containing 0.5 ng/μL THC-d3, THC–OH-d3, CBD-d3, CBN-d3 and 5 ng/μL THC–COOH-d9 in MeOH was prepared. Ethyl acetate (EtOAc) in analytical grade was purchased from Roth (Arlesheim, Switzerland). Analytical grade water, MeOH and acetonitrile (ACN) were obtained from Macherey-Nagel AG (Oensingen, Switzerland). *N*-Methyl-*N*-(trimethylsilyl)trifluoroacetamide (MSTFA) synthesis grade and β-glucuronidase were purchased from Sigma Aldrich (Buchs, Switzerland). Potassium dihydrogen phosphate in analytical grade, sodium hydroxide pellets and glacial acetic acid (100%) were obtained from Merck (Zug, Switzerland). Certified quality control (QC) blood lyophilisate with a concentration of 2.92 ng/mL THC, 43.1 ng/mL THC–COOH and 2.25 ng/mL THC–OH after reconstitution was obtained from ACQ Science (Rottenburg-Hailfingen, Germany).

2.2. Study design

The study volunteer was a 37 years old female weighing 69 kg (BMI 23.6), non-smoker, who had not consumed any cannabinoid containing products prior to the study.

In the first experiment (hereinafter referred to as naive-smoker experiment), the blood and urine cannabinoid concentrations of the naive volunteer after a single CBD joint were investigated. A CBD joint was smoked over a period of approximately 10 min. The smoke was deeply inhaled and held in the lungs for a few seconds prior to exhaling. Blood samples were taken before smoking, immediately after finishing the joint and then approximately every ten minutes until one hour after the start of smoking. Afterwards, blood samples were collected approximately every 30 min until 4 h after the start of smoking. Blood samples were drawn using a peripheral venous catheter and collected in Vacutainer® (Becton Dickinson AG, Allschwil, Switzerland) tubes with sodium fluoride/potassium oxalate stabilization. Urine samples were collected prior to the start of smoking and from every void for 20 h after smoking. Approximately 30 min after smoke cessation, a DrugWipe 6S (Securetec Detektions-Systeme AG, Neubiberg, Germany) test with oral fluid was done.

The blood and urine cannabinoid concentrations of a modeled chronic user after a CBD joint were investigated. For this, a 10-day smoking period was commenced 35 days after the naive-smoker experiment, at which point the participant was again considered naive. During this period, the participant smoked 2 joints each day, giving a total of 20 joints. Before and after smoking the first daily joint a urine sample was collected. On 5 days during the 10-day smoking-period, blood samples were collected shortly before and approximately 30 min after smoking the second daily joint (approximately 6–8 h after the first joint). When smoking the last joint of the 10-day smoking period the second experiment was conducted (hereinafter referred to as chronic-smoker experiment) and the same sampling scheme as in the naive-smoker experiment was repeated (excluding the DrugWipe test).

All blood and urine samples were stored at 4 °C until analysis. Blood samples were analyzed within one week and urine samples within two weeks after sampling.

2.3. Cannabis material

For the naive-smoker experiment, the CBD cannabis product "Indoor Haze" was purchased from Hempner (Hanfkultur GmbH, Fisibach, Switzerland). For the chronic-smoker experiment, a mixed sample of CBD cannabis, obtained from seizures in local CBD-shops, was used, as "Indoor Haze" was no longer available for purchasing. The mixed sample was finely homogenized before splitting into single portions. The cannabinoid content of the plant material was measured in-house using a validated GC–MS method for cannabis plant material to confirm the declared content and to determine the content of the mixed sample. THC-A and CBD-A were converted directly to THC and CBD in the injector of the GC, giving THC_{total} and CBD_{total}. The declared content of "Indoor Haze" was 23% CBD and 0.8% THC. This was confirmed with measured concentrations of 23.5% CBD_{total} and 0.94% THC_{total}. The cannabinoid content of the mixed sample was measured as 17% CBD_{total} and 0.8% THC_{total}.

Joints contained 500 mg tobacco and 200 mg CBD cannabis, which corresponded to 47 and 34 mg CBD and 1.9 and 1.6 mg THC in the naive-smoker and chronic-smoker experiments, respectively.

2.4. Sample preparation

Blood samples were allowed to reach room temperature and 0.50 (±0.01) g were transferred to a centrifuge tube, diluted with 0.5 mL water and spiked with 6 μL internal standard mix. Protein precipitation was done by addition of 2 mL ACN while vortexing. Samples were centrifuged for 5 min at 3000 rpm. The supernatant was transferred to a headspace vial, further diluted with 3 mL of water and added to the autosampler for the automated sample preparation. Every 15 samples, a QC sample with 0.5 g of reconstituted control blood lypophilisate was prepared. CBN and CBD (different Lot. number than for the calibration) were added to the control blood prior to sample preparation, resulting in a final sample concentration of 0.6 ng/mL for both analytes.

Urine samples were allowed to reach room temperature. A centrifuge tube was filled with 1.00 (±0.01) g of urine and spiked with 6 μL internal standard mix to which 1 mL phosphate buffer (0.1 M, ph 6.8) and 25 μL β-glucuronidase (E. coli, 500 units) were added. The sample was enzymatically hydrolyzed for 16 h at 37 °C. After cooling to room temperature, 2 mL ACN were added while vortexing. After centrifugation, the supernatant was transferred to a headspace vial, diluted with 3 mL water, and placed on the autosampler for the automated sample preparation.

2.5. Automatic online sample preparation

Solid phase extraction (SPE) was performed online on a MultiPurposeSampler 2 (Gerstel GmbH, Mülheim an der Ruhr, Germany) using Chromabond C18 ec polypropylene cartridges (1 mL/100 mg, Macherey-Nagel, Oensingen, Switzerland). The SPE cartridge was conditioned with 2 mL MeOH, followed by 2 mL water, and finally with 1 mL 0.1 M acetic acid. The SPE cartridge was loaded with 5 mL of the pre-treated sample. The cartridge was washed with 1 mL 0.1 M acetic acid, 1 mL ACN:water 2:3 and was dried under nitrogen. The cartridge was eluted with 1.4 mL ACN.

The eluate was evaporated to dryness and 30 μL *N*-methyl-*N*-(trimethylsilyl) trifluoroacetamide and 20 μL dry EtOAc were added. Sample derivatization was carried out at 90 °C for 17 min.

2.6. GC–MS method

A Trace GC Ultra gas chromatograph was coupled to a TSQ Quantum mass spectrometer (both Thermo Fischer Scientific, Waltham, USA). Separation was achieved with an Optima 5 MS column (30 m, 0.25 mm ID, 0.25 μm film thickness, Macherey-Nagel, Oensingen, Switzerland).

Analysis was done by injecting 2 μL of sample in PTV splitless mode with an injection temperature of 70 °C increased to 250 °C with 14.5 °C/s. The carrier gas (He) flow through the column was 1.5 mL/min. The GC temperature program was as follows: initial temperature 70 °C held for 0.5 min, increased to 200 °C at 80 °C/min, increased to 300 °C at 10 °C/min and held for 3 min for a total run time of 15 min. The transfer line temperature was set to 250 °C. The selected reaction monitoring (SRM) conditions and retention times are shown in Table 1. The filament emission current was set to 50 μA with a source temperature of 250 °C. Data acquisition, instrument control and data evaluation were performed with Thermo Xcalibur (version 2.1.0.1140, Thermo Fischer Scientific, Waltham, USA).

2.7. Urine immunoassay testing

All urine samples were tested with the Indiko™ Plus immunoassay (Cannabinoids Multi Level THC CEDIA test kit, Thermo Fischer Scientific) using a cut-off of 50 ng/mL for THC–COOH prior to GC–MS/MS analysis.

2.8. Urine creatinine normalization

To better compare the urine samples, the analyte concentrations were normalized to the creatinine content of the urine. Creatinine normalization was done as described by Cone et al. [15]:

$$Conc._{Norm} = \frac{Conc._{Sample}}{CR_{Sample}} * CR_{Ref} \quad (1)$$

where CR_{Ref} is a reference concentration chosen as 100 mg/dL, which is close to the mean of all samples of the volunteer of 92 mg/dL, $Conc._{Sample}$ is the analyte concentration [ng/mL] in the sample, CR_{Sample} is the creatinine concentration [mg/dL] in the sample and $Conc._{Norm}$ is the creatinine normalized concentration [ng/mL] of the sample.

3. Results and discussion

3.1. Validation parameters

The method was validated according to the guidelines of the Swiss Society of Legal Medicine and has proven its suitability for forensic–toxicological routine work by successful application in proficiency tests. The validation parameters can be found in Table 2.

3.2. Cannabis rapid screening test

The DrugWipe saliva test yielded a negative result. According to the manufacturer, the DrugWipe 6S test has a detection limit of 5 ng/mL for THC and no cross reactivity with CBD.

3.3. Blood cannabinoid concentrations

The cannabinoid blood concentration profiles of the naive-smoker and chronic-smoker experiments are shown in Fig. 1. The numerical results can be found in the Supplementary material. All time points are given relative to smoking start (t = 0 min).

3.3.1. Naive-smoker experiment

No cannabinoids were detected in the blood sample taken prior to smoking in the naive-smoker experiment. After smoking, a maximum CBD concentration of 45.7 ng/mL was found in the first blood sample at ca. 10 min. The concentrations remained above the LOQ during the entire collection time with a concentration of 0.9 ng/mL at 4.5 h. The highest THC concentration, 2.7 ng/mL, was also found in the first blood sample after smoking. The concentration quickly declined, dropping below the decision cut-off (2.2 ng/mL) in the second blood sample taken at ca. 20 min and below the LOD (0.2 ng/mL) in the fourth sample taken at ca. 40 min. No THC–OH (LOD = 0.2 ng/mL) or THC–COOH (LOD = 2.0 ng/mL) was detected in any sample. CBN was detected in the first blood sample after smoking at a concentration below the LOQ (0.3 ng/mL)

3.3.2. 10-day smoking period

On 5 days of the 10-day smoking period leading up to the chronic-smoker experiment blood samples were collected before and after smoking the second daily joint. The results of these samples are shown in Table 3. Starting from the second day of the 10-day period some CBD could be found in every blood sample taken before smoking with concentrations in the range of ca. 0.4–

Table 1
SRM conditions and analyte retention times.

Analyte	Precursor ion (m/z)	Product ion (m/z)	Retention time (min)	Collision energy (eV)
THC	371.2	289.3	9.0	20
	386.3	371.4		13
THC–OH	371.3	289.2	10.9	15
THC–COOH	371.3	289.2	11.9	15
	488.3	398.3		15
CBN	367.4	295.2	9.7	30
	367.4	310.2		20
CBD	390.2	231.05	8.1	20
	390.2	300.8		10
THC-d3	374.3	292.2	9.0	20
	389.3	374.4		13
THC–OH-d3	374.3	292.3	10.9	15
THC–COOH-d9	380.3	292.2	11.8	15
	497.3	380.4		15
CBN-d3	370.5	295.3	9.7	30
	370.5	310.3		25
CBD-d3	393.07	304.53	8.1	10

Table 2
Validation parameters of the cannabinoids. The method was validated for blood. Selectivity and linearity were verified for urine. Application of the method in routine use has shown that validation parameters obtained for blood are transferrable to urine analysis. Signal to noise ratios (S/N) were used to determine LOD (S/N ≥ 3) and LOQ (S/N ≥ 10) based on of the least abundant transition in fortified blank serum samples with the additional acceptance criteria that the calculated concentration must be within ±20% of the nominal value and imprecision lower than ±20%.The expanded measurement uncertainty was estimated using 50 blood control samples over a one year period.

Analyte	Linearity (ng/mL)	LOD (ng/mL)	LOQ (ng/mL)	Extraction efficiency (%)	Accuracy bias (%)	Expanded measurement uncertainty (%) (2 × CV%)
THC	0.5–20	0.2	0.3	113 ± 5	2.7	18
THC–OH	0.5–20	0.2	0.3	95 ± 2	−1.3	20
THC–COOH	5–200	2.0	3.0	105 ± 7	−10.3	23
CBN	0.5–50	0.2	0.3	90 ± 10	−10.3	41
CBD	0.5–100	0.2	0.3	88 ± 13	−6.7	38

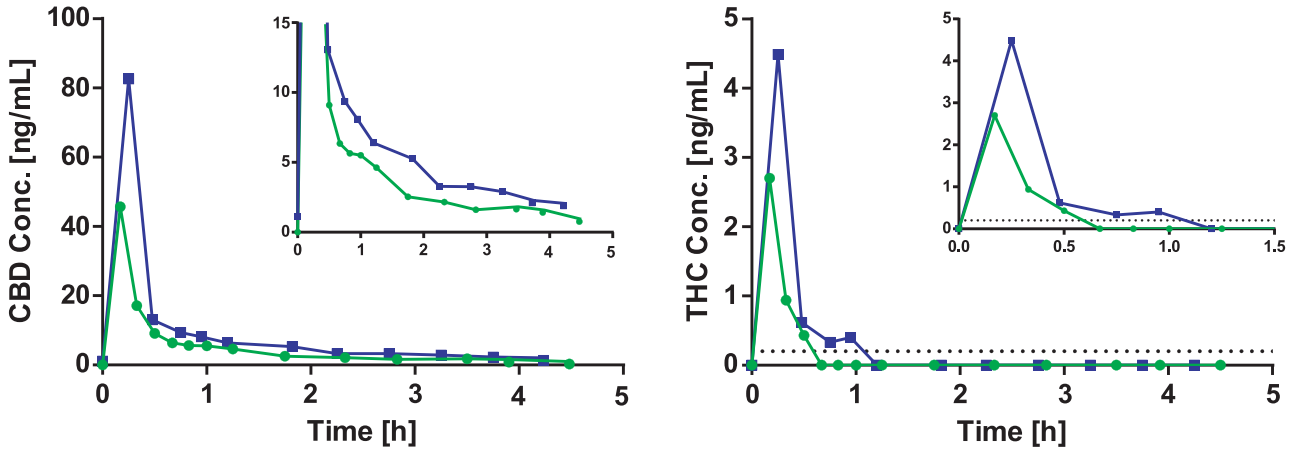

Fig. 1. CBD (left) and THC (right) blood concentrations obtained for the naive-smoker (circles) and chronic-smoker (squares) experiments. The dotted line shows the limit of detection (0.2 ng/mL). The smaller panels within the graphs show enlarged areas of interest.

Table 3
CBD and THC concentrations in the blood samples collected before and after smoking the second daily joint during the 10-day smoking period.

Day	Time between smoking start and sampling (min)	CBD before/after smoking (ng/mL)	THC before/after smoking (ng/mL)
2	20	1.3/13.7	<LOD/0.7
3	25	1.2/15.0	<LOD/0.6
4	30	0.6/11.5	<LOD/0.7
8	30	ca. 0.4/11.7	<LOD/0.5
9	30	0.5/10.5	<LOD/<LOD
Mean	27	0.8/12.5	–/0.5[a]
StD Dev	–	0.4/1.8	–/0.3[a]

[a] Mean and StD Dev calculated with <LOD = 0 ng/mL.

1.3 (mean 0.8 ± 0.4) ng/mL but no trend in the concentration was observed. THC could not be detected in any sample taken directly before smoking. THC was detectable in the samples taken at 20–30 min after smoking in 4 of 5 samples with concentrations between 0.5 and 0.7 ng/mL. The obtained CBD concentrations after smoking were between 10.5 and 15 (mean 12.5 ± 1.8) ng/mL. THC–COOH,THC–OH, and CBN were not detectable in any blood sample collected before or after smoking.

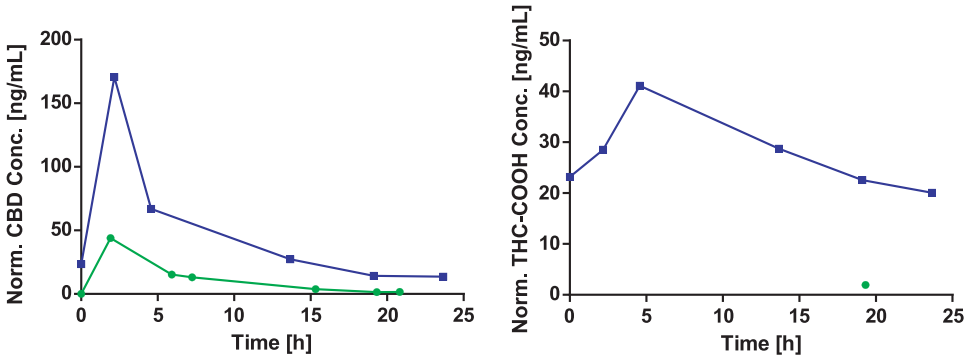

Fig. 2. Creatinine normalized CBD (left) and THC–COOH (right) urine concentrations obtained for the naive-smoker (circles) and chronic-smoker (squares) experiments.

3.3.3. Chronic-smoker experiment

The CBD concentration in the blood sample taken prior to smoking the last joint of the 10-day smoking period and thereby commencing the chronic-smoker experiment was 1.1 ng/mL. No other cannabinoid was detected in this blood sample. After smoking, a maximum CBD concentration of 82.6 ng/mL was found in the first blood sample withdrawn at ca. 15 min. CBD concentration remained above the LOQ for the entire collection period of 4.25 h with a final concentration of 2.1 ng/mL. The highest THC concentration of 4.5 ng/mL was also obtained in the first blood sample after smoking. THC concentrations dropped below the decision cut-off in the second blood sample at ca. 30 min and below the LOD in the fifth sample at ca. 70 min. A THC–OH concentration of ca. 0.3 ng/mL was found in first blood sample. THC–OH concentration in the second sample at ca. 30 min was still above the LOD. No THC–OH could be detected in the following samples. CBN was only detected in the first blood sample after smoking with a concentration of 0.8 ng/mL. THC–COOH could not be detected in any sample.

3.4. Urine cannabinoid concentrations

Urine results of the naive-smoker and chronic-smoker experiments are shown in Fig. 2. All urine concentrations are given as creatinine normalized concentrations. All time points are given relative to smoking start (t =0 min).

3.4.1. Naive-smoker experiment

For the naive-smoker experiment, no cannabinoids were detected in the urine before smoking. A maximum CBD concentration of 44.0 ng/mL was observed in the first urine at 1.9 h. In each subsequent urine the concentrations continuously decreased. A CBD concentration of 1.7 ng/mL was found in the last urine sample given at 20.8 h. THC–COOH was only detectable in one urine sample, 19.3 h after smoking, at a concentration of 1.9 ng/mL (non-normalized ca. 3.6 ng/mL). This sample showed a much higher (factor three) creatinine concentration than any other urine sample, suggesting concentrated urine. This is likely the reason THC–COOH could be detected in only this sample. No THC was found in any urine sample.

3.4.2. 10-day smoking period

The results of the urine samples taken before and after smoking the first joint each day during the 10-day smoking period are shown in Fig. 3. CBD could be detected in every sample with a maximum concentration of 219 ng/mL. A maximum THC–COOH concentration of 42.5 ng/mL was found on the eighth day. The samples marked with a large circle in Fig. 3 gave positive results with the immunoassay test.

3.4.3. Chronic-smoker experiment

The urine sample taken prior to smoking the last joint of the 10-day period and thereby commencing the chronic-smoker experiment showed CBD and THC–COOH concentrations of 23.4–23.2 ng/mL, respectively. After smoking, a maximum CBD concentration of 171 ng/mL was found at 2.2 h. The highest THC–COOH concentration was found at 4.6 h with 41.1 ng/mL. No THC was found in any sample.

4. Discussion

The concentration profiles of THC and CBD were similar to each other as well as being similar in both the naive-smoker and chronic-smoker experiments. Both CBD and THC showed a sharp increase in concentration immediately after smoking, followed by a rapid decrease from distribution of the cannabinoids into the tissues, and finally a very slow elimination from blood. The observed biphasic pharmacokinetics of CBD and THC are in line with the profiles described in the literature [16,17].

In Switzerland, the limit according to the relevant ordinance [14] for THC in whole blood is 1.5 ng/mL for driving. At blood levels $\geq$1.5 ng/mL the driver is defined as momentarily unfit for driving, regardless of whether or not the accused presents typical cannabis-related symptoms or shows inappropriate driving maneuvers. Considering the harmonized imprecision used in Switzerland of 30% results in a decision threshold value of 2.2 ng/mL. The THC blood concentrations of the participant rose above this limit in both experiments, after which the concentrations dropped below 2.2 ng/mL at around 30 min after smoking start. Thus, individuals smoking CBD cigarettes or joints directly prior to or during driving could be unfit for driving according to the Swiss Road Traffic Act and risk losing their driver's license if they are subjected to a control by the police. Assuming similar concentration-time profiles as seen in this study, only blood samples collected shortly after CBD consumption could result in THC blood levels above the legal limit. Usually, there is some time between being stopped by the police and the collection of blood specimens. Therefore, the risk of losing the driver's license due to smoking a CBD joint is relatively low. However, as blood levels can exceed the limit for THC, we fully support the recommendation by the Swiss Federal Office of Public Health to not smoke CBD joints prior to driving.

THC did not show any accumulation during the 10-day smoking period. Smoking experiments comparing cannabinoid concentrations in whole blood of occasional and regular cannabis smokers showed significant THC accumulation in regular cannabis users [18]. However, it seems that the consumed amount of THC in the present study was too low to give detectable accumulation of THC. If more or stronger joints were smoked per day or if smoking would be continued for a longer time period, it cannot be ruled out that some

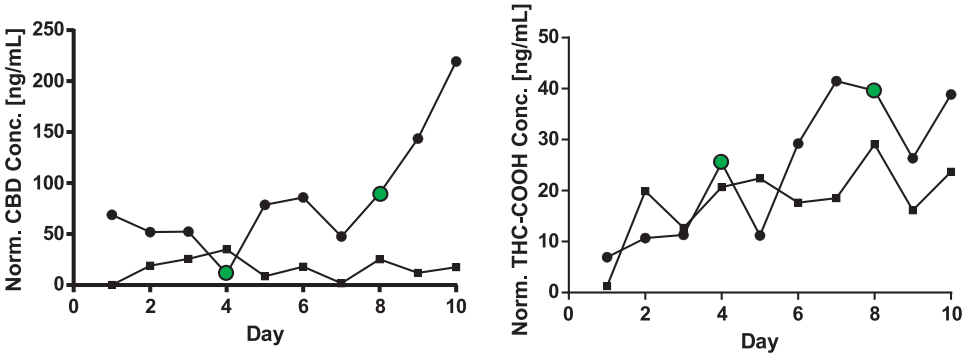

Fig. 3. Creatinine normalized CBD (left) and THC–COOH (right) urine concentrations obtained in the urine samples taken before (squares) and after (circles) smoking the first joint of each day during the 10-day smoking period. The large circles show samples which tested positive using the immunological test with a THC–COOH cut-off of 50 ng/mL.

accumulation might occur. Low levels of CBD were found in blood samples taken prior to smoking the second daily joint during the 10-day smoking period. However, there was no continuous CBD concentration increase during this period, although such a trend might be obscured by the variation in sampling times after smoking.

As is visible in Fig. 3, there seems to be a trend of increasing urinary THC–COOH concentrations during the 10-day smoking period. This could be relevant for cannabinoid testing in urine e.g. in cannabis abstinence testing. Urine cannabinoid concentrations are difficult to interpret and a conclusive differentiation between CBD- or THC-rich cannabis consumption might not always be possible.

The mayor limitation of this study is that it was only conducted with a single person. Therefore, the presented results should be carefully interpreted as inter-individual differences can be expected. Studies with more participants, over a longer time period and with a more frequent sampling scheme are needed to give more concrete recommendations for driving fitness and for providing a basis to differentiate between licit and illicit cannabis consumption. Additionally, sampling frequency during the adsorption phase was most likely too low to capture the maximal blood concentration. Especially in the minutes directly after smoking start, more frequent sampling would have been needed to describe the initial phase sufficiently.

5. Conclusion

The presented case study shows that immediately following smoking of a CBD rich joint (THC below 1%), THC concentrations above the legal limit for driving in Switzerland can be reached. Therefore, CBD smokers should refrain from driving for some hours after smoking. No accumulation of THC was seen when smoking 2 joints per day over a 10-day period.

6. CRediT authorship contribution statement

Ulf Meier: Conceptualization, Investigation, Writing – Original Draft, Visualization. **Franz Dussy:** Conceptualization, Investigation, Writing – Review & Editing. **Eva Scheurer:** Writing – Review & Editing. **Katja Mercer-Chalmers-Bender:** Writing – Review & Editing. **Sarah Hangartner:** Validation, Conceptualization, Methodology, Investigation, Writing – Review & Editing, Project Administration.

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Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at https://doi.org/10.1016/j.forsciint.2018.08.009.

References

[1] R. Ramer, S. Fischer, M. Haustein, K. Manda, B. Hinz, Cannabinoids inhibit angiogenic capacities of endothelial cells via release of tissue inhibitor of matrix metalloproteinases-1 from lung cancer cells, Biochem. Pharmacol. 91 (2) (2014) 202–216.
[2] A. Ligresti, A.S. Moriello, K. Starowicz, et al., Antitumor activity of plant cannabinoids with emphasis on the effect of cannabidiol on human breast carcinoma, J. Pharmacol. Exp. Ther. 318 (3) (2006) 1375–1387.
[3] O. Devinsky, J.H. Cross, L. Laux, et al., Trial of cannabidiol for drug-resistant seizures in the dravet syndrome, N. Engl. J. Med. 376 (21) (2017) 2011–2020.
[4] O. Devinsky, E. Marsh, D. Friedman, et al., Cannabidiol in patients with treatment-resistant epilepsy: an open-label interventional trial, Lancet Neurol. 15 (3) (2016) 270–278.
[5] E.C. Rosenberg, R.W. Tsien, B.J. Whalley, O. Devinsky, Cannabinoids and epilepsy, Neurotherapeutics 12 (4) (2015) 747–768.
[6] T. Iuvone, G. Esposito, D. De Filippis, C. Scuderi, L. Steardo, Cannabidiol: a promising drug for neurodegenerative disorders? CNS Neurosci. Ther. 15 (1) (2009) 65–75.
[7] A.W. Zuardi, I. Shirakawa, E. Finkelfarb, I.G. Karniol, Action of cannabidiol on the anxiety and other effects produced by delta 9-THC in normal subjects, Psychopharmacology (Berl) 76 (3) (1982) 245–250.
[8] R.B. Laprairie, A.M. Bagher, M.E. Kelly, E.M. Denovan-Wright, Cannabidiol is a negative allosteric modulator of the cannabinoid CB1 receptor, Br. J. Pharmacol. 172 (20) (2015) 4790–4805.
[9] S. Pisanti, A.M. Malfitano, E. Ciaglia, et al., Cannabidiol: state of the art and new challenges for therapeutic applications, Pharmacol. Ther. 175 (2017) 133–150.
[10] UNODC, World Drug Report 2017, United Nations Publication, 2017.
11 Eidgenössisches Departement des Innern. Betäubungsmittelverzeichnisverordnung. Admin.ch. https://www.admin.ch/opc/de/classified-compilation/20101220/index.html. Published May 2011. (Accessed 22 March 2018).
[12] EMCDDA, Cannabis Legislation in Europe: An Overview, Publications Office of the European Union, Luxembourg, 2017.
[13] BAG, BLV, BLW, Produkte Mit Cannabidiol (cbd) — Überblick Und Vollzugshilfe, Schweizerische Eidgenossenschaft, 2017.
[14] Der Eidgenössische Bundesrat. Verordnung über die Betäubungsmittelkontrolle. Admin.ch. https://www.admin.ch/opc/de/classified-compilation/20101221/index.html. Published Mai 2011. (Accessed 22 March 2018).
[15] E.J. Cone, Y.H. Caplan, F. Moser, T. Robert, M.K. Shelby, D.L. Black, Normalization of urinary drug concentrations with specific gravity and creatinine, J. Anal. Toxicol. 33 (1) (2009) 1–7.
[16] M.A. Huestis, A.H. Sampson, B.J. Holicky, J.E. Henningfield, E.J. Cone, Characterization of the absorption phase of marijuana smoking, Clin. Pharmacol. Ther. 52 (1) (1992) 31–41.
[17] A. Ohlsson, J.E. Lindgren, S. Andersson, S. Agurell, H. Gillespie, L.E. Hollister, Single-dose kinetics of deuterium-labelled cannabidiol in man after smoking and intravenous administration, Biomed. Environ. Mass Spectrom. 13 (2) (1986) 77–83.
[18] M. Fabritius, H. Chtioui, G. Battistella, et al., Comparison of cannabinoid concentrations in oral fluid and whole blood between occasional and regular cannabis smokers prior to and after smoking a cannabis joint, Anal. Bioanal Chem. 405 (30) (2013) 9791–9803.